

Cereal leaf beetle, *Oulema melanopus* (L.) (Coleoptera: Chrysomelidae), attraction to oat plantings of different ages

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ABSTRACT

The cereal leaf beetle (CLB), *Oulema melanopus*, a serious pest in oats, barley and wheat, is a relatively new pest on the west coast of North America. To determine if adults showed a preference for oat stands of different ages, we examined adult and egg densities in four sequentially planted oat stands in the Willamette Valley, Oregon, in 2005 and 2006. Adults moved from earlier to later plantings (from older to younger oats) during the growing season, particularly once the flag leaf had emerged in earlier plantings. In 2005, the seasonal pattern in egg counts tended to match that of adult counts in the first three oat plantings. The egg to adult ratio was greater in the earlier planted (older) oats, particularly the first planting. The egg to adult ratio was more variable in 2006. Adults spent the most physiological time (degree-days) in the second oat planting, and total egg numbers were highest in the second and third plantings. Data suggest that delayed planting as a trap crop management tool for CLB is complex and potentially ineffective.

Key Words: cereal leaf beetle, *Oulema melanopus*, plant age, host attraction, egg production, management

INTRODUCTION

The cereal leaf beetle (CLB), *Oulema melanopus* (L.) (Coleoptera: Chrysomelidae), is a new pest in cereals in the western United States (Rao *et al.* 2003). CLB was first detected in Michigan in 1962 and soon became a serious pest of small grains in the Midwest, the Atlantic States, and eastern Canada. Wheat (*Triticum aestivum* L.), barley (*Hordeum vulgare* L.), and oats (*Avena sativa* L.) were damaged (Haynes and Gage 1981), with yield reductions in oats reaching 30% (Wilson *et al.* 1969) to 48.8% (Merritt and Apple 1969). Incorporation of trichome resistance in experimental wheat varieties reduced CLB numbers, although effective trichome resistance was not present in other grains (Haynes and Gage 1981). Biological control efforts were initiated and, once the gregarious larval parasitoid *Tetrastichus julis* (Walker) (Hymenoptera: Eulophidae) and the egg parasitoid *Anaphes flavipes* (Foerster)

(Hymenoptera: Mymaridae) became established, damaging populations requiring chemical control measures were substantially reduced (Haynes and Gage 1981). CLB was first detected in Oregon and Washington in 1999 (Rao *et al.* 2003), where it caused direct damage to cereal crops. In addition, quarantine restrictions were established on movement of hay and forage from infested counties in Oregon and Washington to neighboring California and Canada due to potential transport of adult CLB in baled straw. As the pest moved into the region, attention was directed towards developing improved monitoring tools (Rao *et al.* 2003) and control tactics. The parasitoid *T. julis* is not widespread in the Pacific Northwest and *A. flavipes* has not yet established.

CLB has one generation per year. Eggs are laid in spring and the larvae develop through four instars by early summer. Pupa-

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tion occurs in earthen cells in the upper 5 cm of soil and adults emerge after two to three weeks after pupation begins. The overwintering adult population dies by mid-June, several weeks before emergence of the first summer adults. New adults feed to a limited extent but do not mate. Adults overwinter within fields if there is a large amount of crop residue, or outside fields in sheltered fencerows and woodlots (Haynes and Gage 1981).

There is no consistent pattern in spring movement of CLB in relation to the age of the host plant. Adults typically move from overwintering sites to grasses and winter wheat and then to emerging spring-planted grains. In winter wheat, which is planted in fall, CLB adults prefer later plantings (Casagrande *et al.* 1977) and more eggs and larvae are found on these plants (Gage

1974). The opposite is found in oats, which are planted in spring. Early plantings of oats have more CLB adults (Casagrande *et al.* 1977), eggs and larvae (Gage 1974). For unknown reasons, adult preference for winter wheat versus spring grains (primarily oats) can change over several years (Casagrande *et al.* 1977), suggesting that relationships between plant species, plant age, and CLB adult preference are not static (Haynes 1973).

In this study we examined the dynamics of CLB adult and egg densities in oat stands of four ages in the Willamette Valley, Oregon. To assess the potential impact of CLB in these stands, we determined the total time adults spent in each stand and the total number of eggs laid. The potential role of host-plant selection behaviours in CLB management strategies is discussed.

MATERIALS AND METHODS

The study was conducted in 2005 and 2006 at the Hyslop Field Laboratory, Oregon State University, in a field of oats (variety Cayuse). This field was adjacent to an oat nursery used by USDA-APHIS and Oregon Department of Agriculture (ODA) for propagating *T. julis*. Four stands of oats (Cayuse) were planted sequentially on 15 March, 5 April, 26 April, 2 June, 2005; and 7 April, 20 April, 10 May and 6 June, 2006. In 2005, the fourth planting was delayed due to the high rainfall in May. In 2006, limited rainfall in late April and May delayed germination of the second planting, so the second and third plantings were irrigated until rains started again in late May. In both years, the four treatments (called planting dates PD1 through PD4 in chronological order) were set up as a randomized complete block design with four replicates. Each plot was 6.85 m wide and approximately 42.5 m long, and consisted of 45 oat rows,

We monitored CLB adult and egg populations in the plots throughout the overwintered adult activity period (mid-April through late June). Sampling began as soon as the first adults were seen in the plots and

continued at weekly intervals until adults were no longer present in the field. In 2005, adults and eggs were counted in ten randomly located subsamples (30.5 cm of row) per block. In 2006, we took five subsamples per block, and low CLB populations necessitated increasing the subsample area for adults to five adjacent rows (30.5 cm sections), and two adjacent rows (30.5 cm sections) for eggs. The sampling regime ended before the emergence of new adults of the next generation. Adult females and males were not differentiated in 2005. In 2006, a representative number of adults was collected from the plots for identification of sex. In 2006, we recorded the plant development stage using the Zadok's Scale (Zadoks *et al.* 1974) to allow for comparisons based on oat plant growth stage.

To calculate the total developmental time adults spent in each of the planting date treatments we transformed the data to a centigrade degree-day (CDD) time scale. Expressing the data on this scale eliminates the effect of variable temperatures on behavior and oviposition, whether seasonal or weekly. Cumulative centigrade degree-days (CCDD) were calculated using temperature

data from the Hyslop weather station, and the CLB development thresholds of 7°C (minimum) and 30°C (maximum) (Guppy and Harcourt 1978). The biofix (start) dates, 6 March in 2005, and 2 April in 2006, for CDD accumulation were based on the date of first adult emergence as predicted by the CLB IPM weather model derived from a synthesis of 10 data sets (IPPC 2009). Adult sample counts were plotted on the CCDD scale, and the graphical method (Southwood 1978) with 20-DD intervals was used to plot the points. The values of cumulative adult degree-days (CADD) per 30.5 cm of row were calculated by summing the area under the curve.

CLB eggs take about two weeks to hatch at spring temperatures in the Willamette Valley. Hence, the same egg could be counted at two or three weekly sampling periods. To calculate the actual total number of eggs laid in each plot over the ovipositional period, egg counts were plotted using the graphical method (Southwood 1978) at 20-DD intervals. The cumulative egg degree-days was divided by the number of degree-days it takes for an egg to hatch (105 CDD; Guppy and Harcourt 1978) to obtain the total number of actual eggs per sample unit (Southwood 1978).

Egg production per adult was calculated by dividing the number of eggs at a given number of accumulated degree-days by the number of 20-DD intervals over which the eggs could have accumulated (5 intervals maximum). This number was then divided by the running average of adults over corresponding time period. This value was not calculated for the fourth planting because of the minimal adult and egg density data.

Adults can move between plots, which raised the question of whether these experimental units were independent. This question was addressed by examining the spatial autocorrelation variogram for egg and adult count data on each sampling date (SAS 9.1).

Repeated-measures analysis was used to

adjust the p-values for temporal autocorrelation present in the within-sampling-date comparisons for differences in egg and adult counts among planting dates. Because counts of adults and eggs increase and then decrease over time we are not interested in the main effects of Julian date and planting date; and effects of planting date are more appropriately analyzed using the cumulative degree-day approach. Count data often fit the Poisson distribution and the comparison of adult counts among planting dates was conducted using PROC Genmod, with parameter options link=log dist=poisson (SAS 9.1). However the low power of this analysis due to the small number of data clusters, and the deviance from the Poisson that occasionally resulted when using the subsample means, led us to analyze the egg count data differently. Egg counts were transformed using the variance-stabilizing transformation developed for CLB egg spatial distributions ($\log_{10}(\text{counts} + 0.13)$) (Logan 1980) and analyzed using repeated measures in PROC Mixed (SAS 9.1). The lack of a similar stabilizing transformation for adults, and the small number of adults in 2006, precluded our using the more powerful mixed model approach for the adult data. For the PROC Mixed and PROC Genmod analysis, we compared planting date means within sampling dates using unadjusted probabilities after first testing for the main effect difference.

To adjust for the statistical problem of modeling zero variance for adult and egg counts where we recorded zero individuals, we dropped these 'planting date by sampling date' entries from the data sets before analysis. Instead, we compared the other counts on this date to zero counts by determining if the 95% confidence limits of the means included zero.

Statistical comparisons of CADD and total eggs among planting dates were performed using ANOVA in PROC GLM with the Tukey adjustment for the number of comparisons (SAS 9.1).

RESULTS

There was minimal spatial autocorrelation in CLB stages among plots in both years. Significant autocorrelation among plots occurred on only one sampling date for adults and one sampling date for egg counts. On all other sampling dates there was either no spatial autocorrelation or the correlation was negative (plots at further distances were more correlated than closer plots). This indicates that the plots were independent from each other and statistics based on the assumption of independent experimental units can be used. It also suggests that significant movement was taking place between the oat plots and adjacent grain fields.

In 2005, overwintered adults appeared after the first two plantings had emerged, and most died by 15 June (Table 1). On each sampling date, CLB adults had a choice of oats of two or more planting date (PD) treatments. No adults were counted in the fourth planting (PD4) on 15 and 24 June, due to the late emergence of oats and declining population of adults, however, a few adults were seen outside the rows sampled. Counts of adults in each planting date treatment generally increased until the following planting date treatment became more attractive, then decreased in the older treatment. Statistical differences among mean counts of adults per planting date treatment occurred on six of eight sampling dates where adults were present (Table 1). Countering this general trend was the decline in adults in the third planting and increase in the second planting during the third week of May (19 May). This was a week of unusually intense rainstorms and we speculate that the adults were seeking refuge in areas of greater plant biomass. At this time the plant height of PD2 was 31.4 cm versus 12.0 cm for PD3. The total number of adults decreased at this time (Table 1) suggesting that some were leaving the oat plantings to seek refuge in other fields or fencerows.

In 2006, sampling started just as the first oat planting emerged. The 2006 adult population was approximately one-tenth that in

2005 (Table 1). The rise and decline in adults over time in each of the plantings was similar to that in 2005, although the trends were not as uniform, perhaps due to the greater variability in the much smaller populations. There were no statistical differences among mean adult counts at $P < 0.05$. However trends in the 2006 count data were similar to 2005 (Table 1). There was a declining trend in adult counts in the oat plantings on the 25 May sampling date. Sampling that week occurred after three days of high rainfall.

When adult population counts over calendar time are transformed to a CDD scale, the sequential movement of adults from early- to late-planted oats through the season can be seen in 2005 and 2006 (Fig. 1a, b). The decline in total adults in the plots each year after several days of heavy rain in late May is notable. It occurred at approximately 310 and 350 CDD in 2005 and 2006, respectively. Expressing the data on a CDD scale also allows calculation of total adult residency time, and total egg numbers, in stands planted at different times. The analysis of CADD in 2005 showed that PD2 had the highest CADD, followed by PD3 and PD1 (Table 2). In 2006, the relative CADDs of the four planting dates were similar to 2005, although the differences were not significant (Table 2). Total CADD in 2005 was 13.88 versus 1.58 in 2006.

On a calendar scale, CLB adults first appeared in the oat plots during the same week in 2005 and 2006. In 2006, the large movement of adults into PD3 coincided with PD1 and PD2 reaching the Zadok's scale of 43 and 33, respectively (boot stage and 3rd node stage) (Table 3). On a degree-day scale (developmental minimum of 7°C) the adults appeared 76.4 CDD earlier in 2006 (Fig. 1). This translates to 12 days at the daily temperatures at this time of year. An even larger difference occurred, 17 days, when a developmental minimum of 9°C (Fulton and Haynes 1975) was used. In 2005, approximately 110 CDD occurred between the time overwintering adults were predicted to emerge, and when they ap-

Table 1.

Weekly mean CLB adult counts in each sequentially planted oat stand.

Planting date	Sampling dates 2005 ¹									
	21-4	29-4	6-5	12-5	19-5	24-5	31-5	9-6	15-6	24-6
15 March	0.03b	0.13	0.15	0.18b	0.10b	0.10b	0.00b	0.00b	0.00	0.00
5 April	0.30a	0.30	0.30	0.28b	0.40a	0.25b	0.20b	0.00b	0.00	0.00
26 April			0.18	0.60a	0.38a	0.58a	1.33a	0.28a	0.00	0.00
2 June									0.00	0.00
Total	0.33	0.43	0.63	1.06	0.88	0.93	1.53	0.28	0.00	0.00

Planting date	Sampling dates 2006									
	21-4	26-4	2-5	10-5	18-5	25-5	1-6	8-6	15-6	23-6
7 April	0.02	0.03	0.03	0.02	0.04	0.02	0.00	0.00	0.00	0.00
20 April			0.02	0.04	0.10	0.01	0.05	0.01	0.01	0.00
10 May						0.06	0.09	0.03	0.03	0.00
6 June										0.00
Total	0.02	0.03	0.05	0.06	0.14	0.08	0.14	0.04	0.04	0.00

¹ Planting date means within year and sampling date with different letters are statistically different at $P < 0.05$.

Table 2.

Cumulative adult degree-days and total egg numbers in the four oat planting dates

Planting date	Cumulative adult degree-days (CADD) ¹	Total number of eggs ²
2005		
15 March	1.53 ± 0.40 b	49.0 ± 5.5 b
5 April	7.78 ± 1.50 a	64.7 ± 6.0 a
26 April	4.49 ± 0.38 b	63.0 ± 2.7 a
2 June	0.00 ± 0.00 c	1.7 ± 0.4 c
Total	13.88	178.4
2006		
7 April	0.37 ± 0.10 a	7.0 ± 1.2 a
20 April	0.65 ± 0.23 a	5.1 ± 0.7 a
10 May	0.54 ± 0.05 a	4.9 ± 0.4 a
6 June	0.00 ± 0.00 b	0.3 ± 0.1 b
Total	1.56	16.3

¹ Means ± SE of PD entries. Within years PDs with different letters are statistically different at $P < 0.05$.

² Total number of eggs per planting date estimated from the area under the curve analysis.

peared in the oat plots. In 2006, this interval was 90 CDD (Fig. 1). Thus there are either errors in the models or CLB adults are on other hosts (winter grains) for several

weeks before migrating into the oats. We did not monitor CLB adults on other hosts early in the season.

In 2005, the temporal pattern in egg

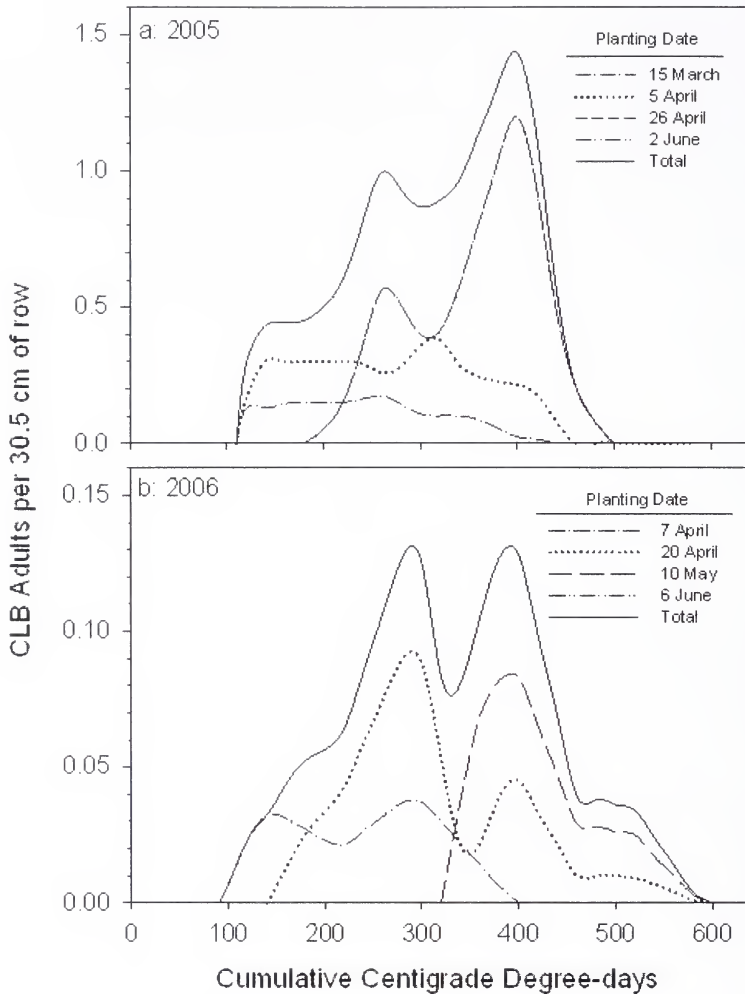


Figure 1. The mean density of CLB adults expressed on a cumulative centigrade degree-day scale, in oat stands of four age classes, i.e., planting dates (PD) 1 – 4: (a) 2005; (b) 2006. The biofix dates were 6 March in 2005, and 2 April in 2006, the dates of first adult emergence predicted by the CLB weather model (IPPC 2009).

density among planting dates was similar to that of the adult populations, with egg counts in each consecutive planting increasing to a peak and then declining as counts increased in the later plantings. There were statistical differences in egg counts on eight of the sampling days (Table 4). The similar egg counts on 19 May are a consequence of the movement of adults out of the younger (smaller) oats, and movement into the older (larger) oats which occurred that week. Quantitative changes in egg density did not always mirror changes in adult density. The egg density in PD2 was only slightly greater than that PD1 (Table 4), even

though the adult population was twice as great in PD2 (Table 1). Similarly, even though the population of adults in PD3 was greater than that in PD2, the number of eggs laid was approximately the same. These relationships between egg and adult counts resulted in the eggs per adult values in PD1 being at times much greater than in PD2 and PD3 (Table 5).

When expressed on a CDD scale, the pattern of egg density over time in the four planting date treatments in 2005 was similar to the calendar day scale (Fig. 2a). The total number of eggs was greater for PD2 and PD3 compared to PD1, while few eggs

Table 3.

Oat plant growth stages (Zadok's scale) at three representative periods during the spring sampling period in 2006

Sampling date	Planting date			
	7 April	20 April	10 May	6 June
2 May	14	11.5	n/a ¹	n/a
1 June	45	33	22.5	n/a
23 June	87	55	33	14

¹ n/a - plants not yet emerged. Zadok's Scale of 14 is a young plant with 4 leaves unfolded; 87 is the hard dough stage of the developing seed.

Table 4.

Weekly mean CLB egg counts in each sequentially planted oat stand

Planting date	Sampling dates 2005 ¹									
	21-4	29-4	6-5	12-5	19-5	24-5	31-5	9-6	15-6	24-6
15 March	0.5 b	16.1	30.0a	28.3b	17.5	16.0b	4.0c	2.0c	0.2c	0.0 b
5 April	2.5 a	18.4	37.4a	34.4a	21.0	22.9a	11.0b	3.9b	1.5b	0.1b
26 April		0.0	4.5b	9.6c	19.3	23.6a	40.3a	30.1a	8.7a	0.3b
2 June									0.4c	2.0a

Planting date	Sampling dates 2006 ¹									
	21-4	26-4	2-5	10-5	18-5	25-5	1-6	8-6	15-6	23-6
7 April	0.3	1.9	2.1a	2.6	3.7	2.2	1.0	0.1b	0.4b	0.0 b
20 April			0.2b	0.9	3.7	1.9	1.6	0.1b	0.2b	0.1b
10 May						2.3	1.8	0.9a	2.5a	1.0a
6 June										0.3b

¹ Planting date means within year and sampling date with different letters are statistically different at $P < 0.05$.

Table 5.

Mean CLB eggs per adult at 100 CDD intervals during the adult activity period¹

Planting date	Accumulative CDD from overwintering adult emergence					
	160	260	360	460	560	
2005						
15 March		33.1	38.2	23.0	16.3	n/a ²
5 April		13.2	24.2	13.5	4.6	n/a
26 April		--	21.5	9.5	7.1	11.0
2006						
7 April		22.1	23.7	13.1	2.1	n/a
20 April		26.9	12.0	6.1	0.6	2.4
10 May		--	--	20.1	3.1	15.3

¹ Number of eggs from Fig. 2 divided by the running average of adults from the previous five 20 CDD periods in Fig. 1 (adults that could have laid those eggs).

² n/a - adults not found in plots

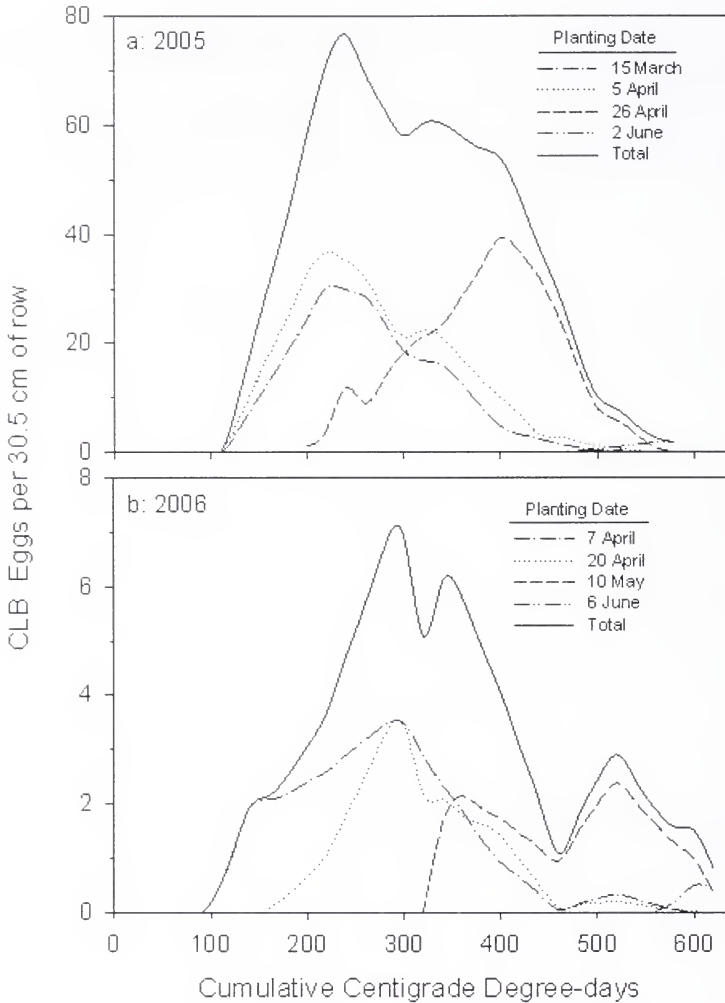


Figure 2. The mean density of CLB eggs expressed on a cumulative centigrade degree-day scale, in oat stands of four age classes, i.e., planting dates (PD) 1 – 4: (a) 2005; (b) 2006. The biofix dates were 6 March in 2005, and 2 April in 2006, the dates of first adult emergence predicted by the CLB weather model (IPPC 2009).

were laid in PD4 (Table 2).

In 2006, the temporal pattern of egg counts did not follow adult densities as well as in 2005. The sequential increase and decrease in the three planting dates was not as uniform. There were significant differences among planting dates in egg counts on four sampling days, mostly at the end of the CLB activity period when PD3 counts were greater than those in the other planting dates (Table 4). The movement of adults from the younger (smaller) oats during the rains of the week of 25 May disrupted the sequential pattern of egg increases in the

younger oats. In 2006, the relative difference among the first three planting date treatments in the eggs per adult value was variable (Table 5), reflecting the lack of correlation between adults and egg counts.

When 2006 egg densities are plotted on a CDD scale (Fig. 2b), the most noticeable difference is that the single peak in total egg density separates into two peaks that correspond to the peaks in total adults (Fig. 1b). There were no statistical differences in the total number of eggs in the first three planting date treatments (Table 2).

DISCUSSION

When CLB adults have a choice of oat stands of different age they tend to prefer younger plants. Overwintered adults move into newly emerged oats after spending time on other hosts, and adult populations increase over time. Populations on older oats decline at the time adult counts are increasing in younger oats. This behaviour is particularly evident once the older oats reach the flag leaf stage. The age of oat leaves affects CLB oviposition, with a dramatic decline in oviposition occurring when a plant approaches the flag leaf stage (GDH unpublished data). This is in contrast to the work of Casagrande *et al.* (1977) that found higher adult populations in older (earlier planted) oats. This contrasting information suggests that adult preference for young plants is not static, and that factors such as weather conditions and the relative differences in plant maturity and size can influence host preference.

There were some differences in the data between 2005 and 2006, in part due to the approximately 10-fold smaller CLB populations in 2006. The 45% parasitism rate of late season larvae by *T. julis* in 2005 (GDH unpublished data) probably accounts for the population decline in 2006. Samples in 2006 contained 100% parasitized CLB larvae after the second generation of *T. julis* (R. Worth unpublished data). One difference between 2005 and 2006 that may have influenced adult and egg counts is the spring drought in 2006. While we used irrigation to get the second and third plantings germinated and established, these later planted stands were probably under greater water stress than the deeper rooted first planting. CLB adults may have been responding to a possible difference in plant water status.

Observations during greenhouse studies showed that many CLB adults leave the oat plants and collect on cage sides between 1000 h and 1600 to 1700 h (unpublished), and the present study documented periodic disappearances of a portion of the within-field population to other habitats. These

observations suggest that CLB adults are moving within and between fields on a regular basis and can respond to changing host plant and environmental cues.

The eggs per adult values calculated from the CDD data are much greater than the eggs per female obtained from laboratory cage experiments. Laboratory data ranged from 8.7 to 12.2 eggs per day at 26.7 °C (Wellso *et al.* 1973). Eight to 18 eggs per day were laid over the first half of post-aestival adult life (Wellso *et al.* 1975). The data from the present study, greenhouse observations, and other studies (Gutierrez *et al.* 1974, Casagrande *et al.* 1977) indicate that CLB adults move frequently within and between fields, and raise the possibility that a significant portion of the adults were not on the oat plants during our mid-day sampling.

In 2005, the impact of CLB was greatest on the second and third oat plantings. More cumulative adult degree-days were recorded in the second planting, and the highest total egg numbers occurred in PD2 and PD3. The fourth planting was minimally affected by CLB due to its late emergence in relation to adult phenology. Similar trends were found in 2006.

While CLB adults were attracted to younger oat plants, preference was not absolute. The variation in adult attraction and egg production in stands of different ages is in part because older oats are not a uniform resource for adults or their developing offspring. Older oats are actually a composite of both young and old leaves, so adult CLB can find young leaves in an older oat stand. In a greenhouse study, the majority of eggs were laid on the softer older leaves, or young tiller leaves of older oat plants (unpublished).

This study suggests that using oat plantings of different ages as a trap crop to help control damage for CLB infestations will be unpredictable and potentially unprofitable. Damage to the flag leaf causes the greatest loss in yield (Yoshida 1972) so the optimum timing of consecutively planted stands

to draw CLB adults away from the primary crop will need to be modeled using input from oat plant growth models, CLB adult preference related to oat phenology, and CLB developmental thresholds. In addition,

late-planted oats are likely to have significantly reduced yields compared to early planted oats (Ciha 1983). Therefore, a late-planted trap crop of oats, even if sprayed to control CLB, will suffer yield loss.

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